Citation:

Binkoski AE, Kris-Etherton PM, Wilson TA, Mountain ML, Nicolosi RJ. Balance of unsaturated fatty acids is important to a cholesterol-lowering diet: Comparison of mid-oleic sunflower oil and olive oil on cardiovascular disease risk factors. *J Am Diet Assoc.* 2005 Jul; 105(7): 1,080-1,086.

PubMed ID: 15983524

Study Design:

Randomized crossover trial.

Class:

A - Click here for explanation of classification scheme.

Research Design and Implementation Rating:

NEUTRAL: See Research Design and Implementation Criteria Checklist below.

Research Purpose:

To evaluate the effects of a *trans* fat-free monounsaturated fatty acid-rich vegetable oil that is a good source of polyunsaturated fatty acids and low in saturated fatty acids on lipid and lipoprotein levels and measures of oxidative stress.

Inclusion Criteria:

- Serum LDL cholesterol between 40th and 90th percentile and high-density lipoprotein (HDL) cholesterol levels between the 25th and 90th percentile for age, race and sex according to Third National Health and Nutrition Examination Survey data
- Triglyceride levels less than 3.95mmol per L
- In good health
- The study was conducted in accordance with the guidelines of the Pennsylvania State University Institutional Review Board, and all subjects gave written informed consent.

Exclusion Criteria:

- Medical condition or history of chronic disease
- Used cholesterol-lowering medication
- Had a body mass index more than 30kg/m^2
- Had lost or gained more than 10lb within the past two months
- Had any lifestyle practices (i.e., irregular work schedule, frequent travel, extreme physical activity, heavy alcohol consumption) that would make it difficult to adhere to the restrictions of the study.

Description of Study Protocol:

Recruitment

Subjects were recruited by a formal screening process that included a telephone interview and a brief physical examination.

Design

Randomized three-period crossover trial.

Blinding Used

Double-blinded study.

Intervention

- The experimental diets provided the same amount of carbohydrate, protein and total fat; half of the fat energy in each test diet was provided by olive oil or NuSun sunflower oil
- Experimental diets met the guidelines of a Step 1 diet (30% total fat, less than 1% SFA, and less than 300mg cholesterol) with approximately energy from total fat and 294mg cholesterol per day, and served a base diet which the test fats were added
- The test fats were incorporated in sauces, spreads, baked goods, granola and salad dressings, as well as in the dinner entrees
- The olive oil and NuSun sunflower oil diets were designed to have the same SFA level, whereas MUFA and PUFA levels were not adjusted and reflected the fatty acid composition of the oils used the respective diet
- Monounsaturated fat content varied between the diets (17.2% for the olive oil diet and 14.2% for the NuSun sunflower oil diet) as did polyunsaturated fat content with 4.3% in the olive oil diet compared with 7.7% in the NuSun sunflower oil diet
- NuSun sunflower (National Sunflower Association, Bismarck, ND) is mid–oleic sunflower oil developed by standard hybrid breeding that contains a similar proportion of and substantially greater proportion polyunsaturated fatty acids (PUFAs) and less SFA compared with olive oil.

Fatty-acid Cmposition of the Fats Tested for Their Effect on Lipid and Lipoprotein Levels and Oxidative Stress in 31 Adults 25 to 64 Years Old with Moderate Hypercholesterolemia

Fatty acid	Olive Oil	NuSun Sunflower Oila
←Percentage→		
Total C18:1	69.4	57.3
Total C18:2	14.0	32.3
Total SFA[b]	14.3	9.6

[a] National Sunflower Association, Bismarck, ND.
[b] SFA = saturated fatty acids.

Statistical Analysis

- All data analyses were performed using SAS (version 8.0,1999-2001, SAS Institute, Cary, NC)
- Data were expressed as least squares means ± standard errors
- The mixed model procedure was used to test four main effects of diet, feeding period, and order of diets
- Tukey-Kramer adjusted P values were used to determine statistical differences between diets for each of the following variables: Serum total cholesterol, LDL cholesterol, HDL cholesterol , triglyceride, apolipoprotein A-l and B, ratio of total to HDL cholesterol, ratio of

LDL cholesterol to HDL cholesterol, lag time, rate of oxidation, total dienes, lipid hydroperoxides, and α -tocopherol
- P-values less than 0.05 were considered significant.

Data Collection Summary:

Timing of Measurements

- Subjects consumed each diet for four weeks followed by a two-week compliance break between diet periods, during which time subjects consumed their habitual diets
- Subjects were weighed daily between Monday and Friday to assure that weight was maintained
- Blood samples were collected in the morning after a 12-hour fast on two consecutive days at the end of each diet period by nurses at The Pennsylvania State University General Clinical Research Center.

Dependent Variables

- Blood was collected into vacutainer tubes (VWR Scientific Products, West Chester, PA) containing SST gel and clot activator for tubes used for serum collection and containing anticoagulant, sodium ethylene diamine tetraacetic acid for tubes used for plasma collection
- Each of the following measurements were completed using established methodological procedures: Lipids and lipoproteins, LDL oxidation, plasma lipid hydroperoxide, plasma LDL tocopherol, total cholesterol (mmol per L), low-density lipoprotein cholesterol (mmol per L), high-density lipoprotein (mmol per L), triglyceride (mmol per L), total cholesterol and high-density lipoprotein cholesterol (mmol per L).

Independent Variables

- The experimental diets provided the same amount of carbohydrate, protein, and total fat; half of the fat energy in each test diet was provided by olive oil or NuSun sunflower oil
- Experimental diets met the guidelines of a Step 1 diet (30% total fat, less than 1% SFA, and less than 300mg cholesterol) with approximately energy from total fat and 294mg cholesterol per day, served a base diet on which the test fats were added
- Every subject consumed each experimental diet in a random, balanced order sequence
- All meals were provided and subjects were required to eat one meal (breakfast or dinner) on a weekday at the Pennsylvania State University Metabolic Diet Study Center
- The other two weekday meals and weekend meals were packed for consumption at a time and place of convenience
- Nonstudy foods and beverages were not permitted with the exception of nonenergy-containing seasonings and beverages
- Compliance was monitored by body weight measurements and a dietary assessment questionnaire administered daily.

Description of Actual Data Sample:

- *Initial N:* 31
- *Attrition (final N):* 31; 12 males, 19 females

- *Age:* 25 to 34 years
- *Location:* University Park, PA.

Summary of Results:

Key Findings

- The NuSun sunflower oil diet decreased both total and LDL cholesterol levels, compared with the average American diet and the olive oil diet
- The NuSun sunflower oil diet significantly reduced total and LDL cholesterol levels, as well as apolipoprotein A-1 levels compared with the average American diet ($P<0.001$, $P=0.0006$ and $P=0.0004$, respectively)
- There was no effect of the olive oil diet compared with the average American diet
- Total cholesterol decreased 4.7% and LDL cholesterol decreased 5.8% on the NuSun sunflower oil diet vs. the average American diet
- There was no effect of the experimental diets on triglyceride levels, rate of oxidation, total dienes, lipid hydroperoxides or α-tocopherol
- Lag time was the longest following the olive oil diet and shortest following the NuSun sunflower oil diet.

Baseline Characteristics of Subjects

Baseline Characteristics of Subjects in a Controlled Feeding Study to Test the Effect of Fatty Acid Composition on Lipid and Lipoprotein Levels and Oxidative Stress (N=31)	
Age (y)	46.2±0.8
Body mass index[b]	26.1±0.3
Total cholesterol (mmol per L)[c]	5.69±0.05
Low-density lipoprotein cholesterol (mmol per L)[c]	3.70±0.0
High-density lipoprotein cholesterol (mmol per L)[c]	1.41±0.02
Triglyceride (mmol per L)[d]	1.30±0.05
Total cholesterol/High-density lipoprotein cholesterol (mmol per L)[c]	4.2±0.1

[a] SE = standard error.

[b] Calculated as kg/mg^2.

[c] To convert mmol per L cholesterol to mg per dL, multiply mmol per L by 38.7. To convert mg per dL cholesterol to mmol per L, multiply mg per dL by 0.026. Cholesterol of 5.00mmol per L = 193mg per dL.

[d] To convert mmol per L triglyceride to mg per dL, multiply mmol per L by 88.6. To convert mg per dL triglyceride to mmol per L, multiply mg per dL by 0.0113. Triglyceride of 1.80mmol per L = 159mg per dL.

The experimental diets provided the same amount of carbohydrate, protein and total fat; half of the fat energy in each test diet was provided by olive oil or NuSun sunflower oil. Monounsaturated fat content varied between the diets (17.2% for the olive oil diet and 14.2% for the NuSun sunflower oil diet) as did polyunsaturated fat content with 4.3% PUFA in the olive oil diet compared with 7.7% in the NuSun sunflower oil diet.

Macronutrients, Fatty Acid, Cholesterol, and Fiber Composition of the Experimental Diets Used to Evaluate the Effect of Fatty Acids on Lipid and Lipoprotein Levels and Oxidative Stress in 31 Adults 25 to 64 Years Old with Moderate Hypercholesterolemia

Diet			
Dietary Constituent	**Average American**	**Olive Oil**	**NuSun Sunflower Oila**
Carbohydrate (percentage of energy)	51.9	55.6	55.4
Protein (percentage of energy)	14.2	14.7	14.8
Fat (percentage of energy)	34.0	29.8	29.8
Saturated fatty acid (percentage of energy)	11.2	8.3	7.9
Monounsaturated fatty acid (percentage of energy)	14.9	17.2	14.2
Polyunsaturated fatty acid (percentage of energy)	7.8	4.3	7.7
Cholesterol (mmoL)[bc]	7.86	7.64	7.64
Fiber (g per 1,000kcal)[b]	9.5	13.7	13.7

[a] National Sunflower Association, Bismarck, ND.

[b] Estimated using the NUTRITIONIST V database(N-Squared Computing , San Bruno, CA.

[c] To convert mmol per L cholesterol to mg per dL, multiply mmol per L by 38.7. To convert mg per dL cholesterol to mmol per L, multiply mg per dL by 0.026. Cholesterol of 5.00mmol per L = 193mg per dL.

Experimental diets met the guidelines of a Step 1 diet (30% total fat, less than 1% SFA, and less than 300mg cholesterol) with approximately energy from total fat, and 294mg cholesterol per day and served a base diet which the test fats were added. The test fats were incorporated in sauces, spreads, baked goods, granola and salad dressings, as well as in the dinner entrees.

NuSun[a] Sunflower Oil Diet Sample Menu (1,800kcal) from Controlled-feeding Study Testing the Effect of Fatty Acid Composition on Lipid and Lipoprotein Levels and Oxidative Stress

Meal	**Amount (g)**

Breakfast	
Yoplait[b] 99% fat-free original yogurt, fruit flavor	227
Blueberries, frozen	70
Skim milk	200
NuSun sunflower oil granola	30
All- Bran cereal	20
Lunch	
Whole-wheat bread	50
Healthy Choice[d] deli smoked ham	53
NuSun sunflower oil honey mustard spread	20
Fig Newtons[e] cookies	15
Rold Gold[f] thin twist pretzels	30
Dinner	
Turkey taco	100
Egg yolk	13
Butter	4
NuSun sunflower oil	20
Romaine lettuce	56
Tomato	60
Sweet corn	110
Old El Paso[b] chunky salsa dip	30
Cheddar cheese, shredded	12
Tostitos[f] baked tortilla chips	30
Snack	
Pear halves, canned in extra light syrup	130
Jell-o[e]	90
[a] National Sunflower Association, Bismarck, ND.	
[b] General Mills, Inc,. Minneapolis, MN.	
[c] Kellogg Co. Battle Creek, MI.	
[d] ConAgra Foods, Inc., Omaha ,NE.	

[e] Kraft Foods Inc. Northfield, IL.

[f] PepsiCo, Purchase, NY.

Effect of Experimental Diets with Varying Fatty Acid Compositions on Lipid and Lipoprotein Levels in Men and Women with Moderate Hypercholesterolemia			
	Average American Diet	**Olive Oil Diet**	**NuSun[a] Flower Oil Diet**
Mean ± Standard Error			
Total cholesterol (mmol per L)[b]	5.75±0.14	5.67±0.14	5.47±014**
Low-density lipoprotein cholesterol (mmol per L)[b]	3.76±0.11	3.72±0.11	3.54±0.11*
High-density lipoprotein cholesterol (mmol per L)[b]	1.36±0.6	1.34±0.06	1.32±0.06
Triglyceride (mmol per L)[c]	1.38±0.11	1.28±0.11	1.34±0.11
Total cholesterol (mmol per L)[b]	4.4±0.2	4.4±0.2	4.4±0.2
Total cholesterol/High-density lipoprotein cholesterol (mmol per L)[b]	2.9±0.2	2.9±0.2	2.8±0.2
Apolipoprotein A1 (g per L)	1.57±0.06	1.56±0.06	1.50±0.06***
Apolipoprotein B (g per L)	1.11±0.03	1.08±0.03	1.08±0.03

[a] National Sunflower Association, Bismarck, ND.

[b] To convert mmol per L cholesterol to mg per dL, multiply mmol per L by 38.7. To convert mg per dL cholesterol to mmol per L, multiply mg per dL by 0.026. Cholesterol of 5.00mmol per L = 193mg per dL.

[c] To convert mmol per L triglyceride to mg per dL, multiply mmol per L by 88.6. To convert mg per dL triglyceride to mmol per L, multiply mg per dL by 0.0113. Triglyceride of 1.80mmol per L = 159mg per dL.

* Significantly different from Average American diet at P<0.05.

** significantly different from Olive oil diet at P<0.05.

Effect of Experimental Diets with Varying Fatty Acid Compositions on the Susceptibility of Low-Density Lipoprotein (LDL) to Oxidation in Men and Women with Moderate Hypercholesterolemia			
	Average American Diet	**Olive Oil Diet**	**NuSun[a] Flower Oil Diet**
Mean ± Standard Error			
Lag time (minute)	73.9±3.8	83.4±3.8	67.9±3.8*

Rate of oxidation(nmol per minute per mg protein)	38.1±2.0	34.5±2.0	37.3±2.0
Maximum dienes (nmol per mg protein)	2,318.2±94.7	2,236.3±94.7	2,331.02±94.7
Lipid hydroperoxides (µmol per L)	65.7±1.1	65.9±1.1	65.9±1.1
α -Tocopherol (ng per mcg LDL protein	8.3±0.8	8.2±0.8	8.5±0.8
[a] National Sunflower Association, Bismarck, ND. * Significantly different from Olive oil diet at P<0.05.			

Author Conclusion:

The higher PUFA content appeared to account for the greater total and low-density lipoprotein cholesterol lowering and reduction in lag time of the NuSun sunflower oil diet. However, the fact that there were no differences in the resulting oxidation products suggests there were no adverse effects on low-density lipoprotein oxidation. Since PUFAs are important for cholesterol lowering, foods that replace saturated fatty acids should include a balance of unsaturated fatty acids.

Reviewer Comments:

Relatively small sample size. Diet periods were not equally sized, olive oil and NuSun sunflower oil diets consumed for four weeks, average American diet consumed for two weeks as a wash-out period. The average American diet was not well defined. Sponsored by the National Sunflower Association.

Research Design and Implementation Criteria Checklist: Primary Research

Relevance Questions

1.	Would implementing the studied intervention or procedure (if found successful) result in improved outcomes for the patients/clients/population group? (Not Applicable for some epidemiological studies)	Yes
2.	Did the authors study an outcome (dependent variable) or topic that the patients/clients/population group would care about?	Yes
3.	Is the focus of the intervention or procedure (independent variable) or topic of study a common issue of concern to nutrition or dietetics practice?	Yes
4.	Is the intervention or procedure feasible? (NA for some epidemiological studies)	Yes

Validity Questions		
1.	**Was the research question clearly stated?**	Yes
1.1.	Was (were) the specific intervention(s) or procedure(s) [independent variable(s)] identified?	Yes
1.2.	Was (were) the outcome(s) [dependent variable(s)] clearly indicated?	Yes
1.3.	Were the target population and setting specified?	Yes
2.	**Was the selection of study subjects/patients free from bias?**	Yes
2.1.	Were inclusion/exclusion criteria specified (e.g., risk, point in disease progression, diagnostic or prognosis criteria), and with sufficient detail and without omitting criteria critical to the study?	Yes
2.2.	Were criteria applied equally to all study groups?	Yes
2.3.	Were health, demographics, and other characteristics of subjects described?	Yes
2.4.	Were the subjects/patients a representative sample of the relevant population?	Yes
3.	**Were study groups comparable?**	Yes
3.1.	Was the method of assigning subjects/patients to groups described and unbiased? (Method of randomization identified if RCT)	???
3.2.	Were distribution of disease status, prognostic factors, and other factors (e.g., demographics) similar across study groups at baseline?	Yes
3.3.	Were concurrent controls used? (Concurrent preferred over historical controls.)	Yes
3.4.	If cohort study or cross-sectional study, were groups comparable on important confounding factors and/or were preexisting differences accounted for by using appropriate adjustments in statistical analysis?	N/A
3.5.	If case control or cross-sectional study, were potential confounding factors comparable for cases and controls? (If case series or trial with subjects serving as own control, this criterion is not applicable. Criterion may not be applicable in some cross-sectional studies.)	N/A
3.6.	If diagnostic test, was there an independent blind comparison with an appropriate reference standard (e.g., "gold standard")?	N/A
4.	**Was method of handling withdrawals described?**	Yes
4.1.	Were follow-up methods described and the same for all groups?	Yes

4.2.	Was the number, characteristics of withdrawals (i.e., dropouts, lost to follow up, attrition rate) and/or response rate (cross-sectional studies) described for each group? (Follow up goal for a strong study is 80%.)	Yes
4.3.	Were all enrolled subjects/patients (in the original sample) accounted for?	Yes
4.4.	Were reasons for withdrawals similar across groups?	N/A
4.5.	If diagnostic test, was decision to perform reference test not dependent on results of test under study?	N/A
5.	**Was blinding used to prevent introduction of bias?**	Yes
5.1.	In intervention study, were subjects, clinicians/practitioners, and investigators blinded to treatment group, as appropriate?	Yes
5.2.	Were data collectors blinded for outcomes assessment? (If outcome is measured using an objective test, such as a lab value, this criterion is assumed to be met.)	Yes
5.3.	In cohort study or cross-sectional study, were measurements of outcomes and risk factors blinded?	N/A
5.4.	In case control study, was case definition explicit and case ascertainment not influenced by exposure status?	N/A
5.5.	In diagnostic study, were test results blinded to patient history and other test results?	N/A
6.	**Were intervention/therapeutic regimens/exposure factor or procedure and any comparison(s) described in detail? Were interveningfactors described?**	???
6.1.	In RCT or other intervention trial, were protocols described for all regimens studied?	Yes
6.2.	In observational study, were interventions, study settings, and clinicians/provider described?	N/A
6.3.	Was the intensity and duration of the intervention or exposure factor sufficient to produce a meaningful effect?	???
6.4.	Was the amount of exposure and, if relevant, subject/patient compliance measured?	Yes
6.5.	Were co-interventions (e.g., ancillary treatments, other therapies) described?	N/A
6.6.	Were extra or unplanned treatments described?	N/A
6.7.	Was the information for 6.4, 6.5, and 6.6 assessed the same way for all groups?	Yes
6.8.	In diagnostic study, were details of test administration and replication sufficient?	N/A
7.	**Were outcomes clearly defined and the measurements valid and reliable?**	Yes

7.1.	Were primary and secondary endpoints described and relevant to the question?	Yes
7.2.	Were nutrition measures appropriate to question and outcomes of concern?	Yes
7.3.	Was the period of follow-up long enough for important outcome(s) to occur?	Yes
7.4.	Were the observations and measurements based on standard, valid, and reliable data collection instruments/tests/procedures?	Yes
7.5.	Was the measurement of effect at an appropriate level of precision?	Yes
7.6.	Were other factors accounted for (measured) that could affect outcomes?	???
7.7.	Were the measurements conducted consistently across groups?	Yes
8.	**Was the statistical analysis appropriate for the study design and type of outcome indicators?**	Yes
8.1.	Were statistical analyses adequately described and the results reported appropriately?	Yes
8.2.	Were correct statistical tests used and assumptions of test not violated?	Yes
8.3.	Were statistics reported with levels of significance and/or confidence intervals?	Yes
8.4.	Was "intent to treat" analysis of outcomes done (and as appropriate, was there an analysis of outcomes for those maximally exposed or a dose-response analysis)?	N/A
8.5.	Were adequate adjustments made for effects of confounding factors that might have affected the outcomes (e.g., multivariate analyses)?	???
8.6.	Was clinical significance as well as statistical significance reported?	Yes
8.7.	If negative findings, was a power calculation reported to address type 2 error?	N/A
9.	**Are conclusions supported by results with biases and limitations taken into consideration?**	???
9.1.	Is there a discussion of findings?	Yes
9.2.	Are biases and study limitations identified and discussed?	No
10.	**Is bias due to study's funding or sponsorship unlikely?**	???
10.1.	Were sources of funding and investigators' affiliations described?	Yes
10.2.	Was the study free from apparent conflict of interest?	???